

Field Capture of Male Melon Flies, *Bactrocera cucurbitae* (Coquillett), in Jackson Traps Baited with Cue-Lure Versus Raspberry Ketone Formate in Hawaii

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Abstract. The melon fly, *Bactrocera cucurbitae* (Coquillett), is an invasive worldwide pest of vegetables, particularly various species of squashes and melons. Because it poses a serious economic threat, many countries operate continuous trapping programs to detect incipient infestations. Detection currently relies on traps baited with a chemical (cue-lure, CL) that is attractive to males of this species. Earlier research suggested that a chemically similar compound (raspberry ketone formate, RKF) is more attractive than CL and thus might improve surveillance efforts. The present study describes the results of field experiments in Hawaii that compare captures of *B. cucurbitae* males in traps baited with (1) CL liquid versus a solid formulation (so-called plugs) of RKF or (2) CL liquid versus RKF liquid. In the first experiment, the traps with CL liquid captured more males than traps baited with RKF plugs in three of the four study sites, with no difference observed between lures at the remaining area. In the second experiment, the traps baited with CL liquid captured more males than traps baited with RKF liquid at two of the four study areas, with no difference observed between lures at the remaining two sites. The discrepancy between earlier studies and the present one highlights the need for additional field testing of RKF to assess its value as a replacement for CL.

Introduction

The melon fly, *Bactrocera cucurbitae* (Coquillett), is a serious global pest of many agriculturally important fruits and vegetables (White and Elson-Harris 1992). Believed to have originated in the Indian subcontinent, *B. cucurbitae* now occurs throughout tropical Asia and the South Pacific islands as well as Mauritius, Réunion, Africa, and Hawaii (White and Elson-Harris 1992, PMP-FFM 2002, Dhillon et al. 2005, De Meyer et al. 2011). Females oviposit in over 100 plant species, mostly in the family Cucurbitaceae, including many commercial crops, such as cucumber (*Cucumis sativus* L.), various squashes (*Cucurbita* spp.), and watermelon (*Citrulus lanatus* (Thunb.) Matsum. & Nakai) (White and Elson-

Harris 1992, Dhillon et al. 2005). Because the species poses an enormous agricultural threat, many countries operate continuous large scale trapping programs to detect incipient infestations. Melon fly males are attracted to raspberry ketone (RK, 4-(4-hydroxyphenyl)-2-butanone), a natural compound occurring in many plant species as well as cue-lure (CL, 4-(p-acetoxyphenyl)-2-butanone), the acetate derivative of RK (Metcalf 1990). Traps baited with CL are used in detection programs worldwide (e.g., Huxham 2004, Gonzalez and Troncoso 2007), including US states, such as California, Florida, and Texas, that are vulnerable to invasion by *B. cucurbitae*. In the US, detection traps are operated with 5–6 ml of CL liquid applied to a cotton wick placed inside a Jackson

trap (IPRFFSP 2006).

Despite its wide usage, CL is recognized as a fairly weak attractant, particularly compared to methyl eugenol (ME), which is a male attractant for other *Bactrocera* species, including the oriental fruit fly, *B. dorsalis* (Hendel) (Jang and Light 1996). For example, in a study conducted in southern California, *B. cucurbitae* and *B. dorsalis* males were released 50 m from traps baited with male lures, and 49% of *B. dorsalis* males were captured (in ME-baited traps) compared to only 5% of *B. cucurbitae* males (in CL-baited traps; Shelly et al. 2010). To improve detection and monitoring programs, Metcalf and Metcalf (1992) investigated the attractiveness of various derivatives of RK and reported that the formate ester of RK, formic acid 4-(3-oxobutyl) phenyl ester (RKF), was more attractive to melon fly males than either RK or CL. In more recent field tests, Casaña-Giner et al. (2003) and Jang et al. (2007) likewise found that captures of *B. cucurbitae* males were generally higher in RKF- than CL-baited traps. The time interval over which this differential attractiveness was evident was dependent on both the lure dose and dispenser type. At higher doses, RKF-baited traps captured more male melon flies than CL-baited traps over longer time intervals, and this difference was more pronounced when lures were embedded in plastic plugs than applied as liquids to cotton wicks. In contrast to these results, Vargas et al. (2010) reported no difference in trap catch of *B. cucurbitae* males baited with CL or RKF embedded in a biologically inert, waxy matrix (SPLAT, Specialized Pheromone and Lure Application Technology (ISCA Tech., Riverside, CA)).

Despite some promising findings, there have been, to our knowledge, no additional studies to compare efficacy of CL and RKF. Here, we present the results of field experiments conducted on Oahu, Hawaii,

that compared trap catch of *B. cucurbitae* males between Jackson traps baited with i) CL liquid applied to cotton wicks versus RKF embedded in plastic plugs or ii) CL liquid versus RKF liquid both applied to cotton wicks.

Materials and Methods

Study sites. Comparisons between CL liquid and RKF plugs were made in November–December 2011 at four sites on Oahu: (1) Waimanalo, a coastal area (< 30 m elevation) on the windward (cloudy, rainy) side of the island, (2) Kapolei, also a low elevation site (< 50 m elevation) but on the leeward (sunny, dry) side of the island, (3) Aloun Farm, approximately 1 km and climatically similar to the preceding site, and (4) Mililani Agricultural Park, a higher elevation area (200 m) in central Oahu. Comparisons between CL liquid and RKF liquid were made in February–April, 2012, at four sites: (1) the Mililani site described above, (2) Aloun farm described above, (3) Haleiwa, in a coffee (*Coffea arabica* L.) field (200 m elevation) 7 km south of the town, and (4) Ewa Beach in mixed residential and agricultural land about 5 km from the Kapolei and Aloun Farm sites. During the study period over all sites, daily minimum temperatures ranged from approximately 16–21°C, and maximum temperatures ranged from approximately 24–28°C, and rainfall amounts varied from approximately 10–15 cm/month for the windward sites (Waimanalo, Haleiwa), 12 cm/month for Mililani, and 5–10 cm/month for the remaining, more leeward sites (Kapolei, Aloun Farm, Ewa Beach). Weather data were derived from Weather-warehouse.com.

The study areas differed in the availability of host plants. Aloun Farm contained large commercial fields of various melons and squashes, while the Mililani site contained small, scattered plots of various host species, including papaya

(*Carica papaya* L.), bitter melon (*Momordica charantia* L.), and ivy gourd (*Coccinia grandis* (L.)). In Haleiwa, the traps were placed in a commercial coffee field adjacent to unmanaged land containing bitter melon and papayas. The remaining sites were a mixture of residential, unmanaged, and small agricultural areas and contained, among other host plants, papaya, bitter melons, tomato (*Solanum lycopersicum* L.), and zucchini (*Cucurbita pepo* L.) plants.

Traps and lures. Jackson traps (Scentry Biologicals Inc., Billings, MT) were used exclusively. These were triangular in shape, white in color, and made of thick, waxed paper (12.7 x 9.5 x 8.4 cm, l:w:h). A removable insert, made of the same waxed paper as the trap body and coated with "stickum", was placed on the bottom of the trap to catch flies. Traps were suspended from tree branches using a metal hanger, with a straight rod that is positioned under the "roof" along the apex of the trap.

All lures—CL liquid, RKF plugs, and RKF liquid—were obtained from Scentry Biologicals Inc. In all tests, we applied a total of 6 ml of the liquid lures (5% naled) to two cotton wicks (each 2.5 cm in length and 2 cm in diameter), which were then placed in a perforated, plastic basket. This basket, in turn, was fastened to the metal hanger and suspended in the middle of the Jackson trap directly above the sticky insert. The RKF plugs also contained 6 g of the lure (5% naled) and were placed singly in plastic baskets, which were then positioned inside the Jackson traps.

Trap placement and servicing. At all study sites, we placed 15 traps baited with CL liquid and 15 traps baited with RKF (either as a liquid or in a plug) in the canopy of non-host trees (1.5–2 m above ground in shaded locations). The most commonly used non-host plants included castor bean (*Ricinus communis* L.), kiawe (*Prosopis pallida* [Humb. & Bonpl.

ex Willd.]), and haole koa (*Leucaena leucocephala* [Lam. De Wit]). The trees selected were separated by a minimum of 30 m. Traps operated for 1 day/week for 6 weeks, with trap placement and collection occurring between 1000–1200 hours. Upon collection, the traps were returned to our laboratory in central Oahu, the sticky inserts were removed for fly counts, and the Jackson traps (with lures still in place) were hung outdoors in the shade for weathering. At the study sites, the same trees were used over the 6-week sampling interval, but the type of trap (CL- or RKF-baited) placed on a particular tree was alternated between successive weeks to control for potential position effects.

Statistical analysis. For all trials, raw data were square root transformed and then subject to a two-way ANOVA, with time (week) and lure type (CL liquid, RKF liquid, or RKF plug) as the main effects. If lure type accounted for a significant amount of variation in trap catch, Tukey's test (test statistic q) was used to compare the catch associated with the two lure types. Analyses were performed using SigmaPlot 11.

Results

Cue-lure liquid versus raspberry ketone formate plugs. At three of the four sampling sites, lure type had a significant effect on trap catch of *B. cucurbitae* males (Table 1; Fig. 1), and in each case the traps baited with CL liquid captured more males than those baited with RKF plugs (Tukey's test: Waimanalo: $q = 5.2$, $P < 0.001$; Kapolei: $q = 6.1$, $P < 0.001$; Mililani: $q = 4.1$, $P = 0.004$). At the remaining site (Aloun Farm), lure type had no significant effect on trap catch. Trap captures varied significantly with time at two of the sites, but at no site was there a significant interaction term between lure type and time (Table 1).

Cue-lure liquid versus raspberry ketone formate liquid. At two of the four

Table 1. Results of two-way ANOVA (using square root transformed data) assessing the effect of cue-lure (CL) liquid vs. raspberry ketone formate (RKF) plugs (lure type) and weeks since start of experiment (time) on the capture of *B. cucurbitae* males in Jackson traps at four sites on Oahu, Hawaii. At each site, 15 traps of each treatment were operated 1 day per week over six consecutive weeks (df values: lure type = 1; time = 5; lure type x time = 5; residual df = 168).

Site	Source of variation	F	P
Aloun Farm	Lure type	2.2	0.14
	Time	5.4	< 0.001
	Lure type x time	0.2	0.96
Mililani	Lure type	8.4	0.004
	Time	3.2	0.009
	Lure type x time	0.3	0.94
Waimanalo	Lure type	13.5	< 0.001
	Time	0.6	0.68
	Lure type x time	0.5	0.78
Kapolei	Lure type	18.6	< 0.001
	Time	1.3	0.28
	Lure type x time	0.2	0.96

sampling sites, lure type had a significant effect on trap catch of *B. cucurbitae* males (Table 2; Fig. 2), and in each case the traps baited with CL liquid captured more males than those baited with RKF liquid (Tukey’s test: Aloun Farm: $q = 3.6$, $P = 0.01$; Haleiwa: $q = 3.5$, $P = 0.01$). Trap captures varied with time at all four sites, but at no site was there a significant interaction term between lure type and time (Table 2).

Discussion

The present results indicate that traps baited with RKF either in a solid matrix (plug) or as a liquid captured fewer *B. cucurbitae* males than traps baited with CL liquid, which represents the standard method of lure presentation in detection programs in California and Florida. This conclusion is based on field sampling conducted on Oahu, Hawaii, over 6-week

intervals at each of eight sites (four with RKF plugs, four with RKF liquid). Traps baited with CL liquid captured significantly more *B. cucurbitae* males in five of the eight intervals, while there was no significant difference in trap catch observed in the remaining three intervals. In no instance did traps baited with RKF catch significantly more flies than traps with CL liquid.

This result contrasts with the earlier studies (Metcalf and Metcalf 1992, Oliver et al. 2002, Casaña-Giner et al. 2003, Jang et al. 2007) that reported higher trap catch of *B. cucurbitae* males in traps baited with RKF than CL. Reasons for this discrepancy are unknown. As outlined by Casaña-Giner et al. (2003) and Jang et al. (2007), although Metcalf and Metcalf (1992) demonstrated the greater attractiveness of RKF relative to CL, there was an apparent reluctance to further in-

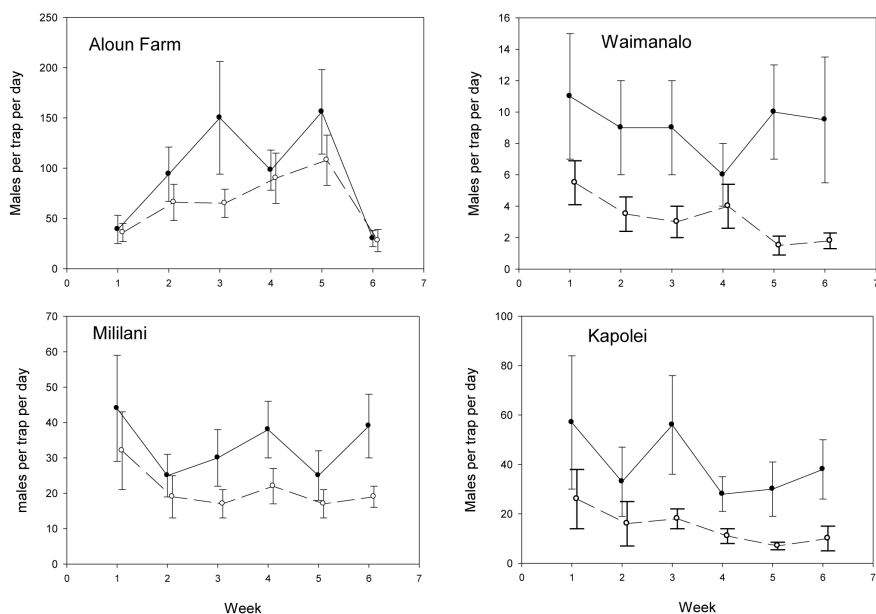


Figure 1. Number of *B. cucurbitae* males captured in Jackson traps baited with cue-lure (CL) liquid (●) versus raspberry ketone formate (RKF) plugs (○) at four study sites on Oahu, Hawaii. At each site, 15 traps of each treatment were operated 1 day per week over 6 consecutive weeks. Symbols represent means (± 1 SE, $n = 15$).

investigate RKF's use in trapping programs owing to concern regarding its hydrolytic instability. That is, the hydrolysis of RKF and its subsequent conversion to RK was expected to occur more rapidly than the hydrolysis of CL to RK. As RK, is less volatile and less attractive than CL (Beroza et al. 1960), more rapid conversion to RK was considered problematic for the adoption of RKF as a bait.

Subsequent work, however, suggested that the rate of hydrolysis of RKF may have been overestimated. Casaña-Giner et al. (2003) found that, under warm, moist field conditions in Hawaii, RKF was fairly stable, with only 15–30% hydrolysis over a 50-day period. Moreover, the rate of conversion of RKF to RK was inversely related to the amount of RKF placed on cotton wicks. At a dose of 1 g (≈ 1 ml), and

presumably at higher doses, Casaña-Giner et al. (2003) concluded that the “hydrolysis threat” does not pose limitations on the use of RKF. The dose of RKF used in the present study (6 g in plug, 6 ml on wick) greatly exceeded those used previously. For example, in their field trapping, Casaña-Giner et al. (2003) applied only 20 mg of lures to cotton wicks, and Jang et al. (2007) compared CL and RKF at doses of 20 mg, 160 mg, or 1 g. The present dose was selected, because it represents the currently accepted dose (in Florida) or is similar to the currently accepted dose (5 ml, California) in large-scale detection programs in the US, and any change in these doses (particularly decreases) are unlikely.

The chemical processes underlying the relative attractiveness of CL, RK,

Table 2. Results of two-way ANOVA (using square root transformed data) assessing the effect of cue-lure (CL) liquid vs. raspberry ketone formate (RKF) liquid (lure type) and weeks since start of experiment (time) on the capture of *B. cucurbitae* males in Jackson traps at four sites on Oahu, Hawaii. At each site, 15 traps of each treatment were operated 1 day per week over six consecutive weeks (df values: lure type = 1; time = 5; lure type x time = 5; residual df = 168).

Site	Source of variation	F	P
Aloun Farm	Lure type	6.4	0.01
	Time	7.2	< 0.001
	Lure type x time	0.2	0.97
Mililani	Lure type	1.8	0.18
	Time	3.2	< 0.001
	Lure type x time	0.7	0.60
Haleiwa	Lure type	6.1	0.01
	Time	21.8	< 0.001
	Lure type x time	0.6	0.71
Ewa Beach	Lure type	0.9	0.36
	Time	5.9	< 0.001
	Lure type x time	0.1	0.98

and RKF, particularly in relation to the hydrolytic conversion of CL and RKF to RK, are not known precisely (Oliver et al. 2002), and description of these processes was beyond the scope of the present study. Regardless of the underlying causes, the present results clearly differ from previous reports, and this discrepancy suggests that further field testing is needed to adequately characterize the relative attractancy of RKF relative to CL.

Acknowledgments

We thank Amanda Ramsey, Scentry Biologicals Inc., for supplying the chemicals and Roger Vargas for helpful comments on an earlier draft.

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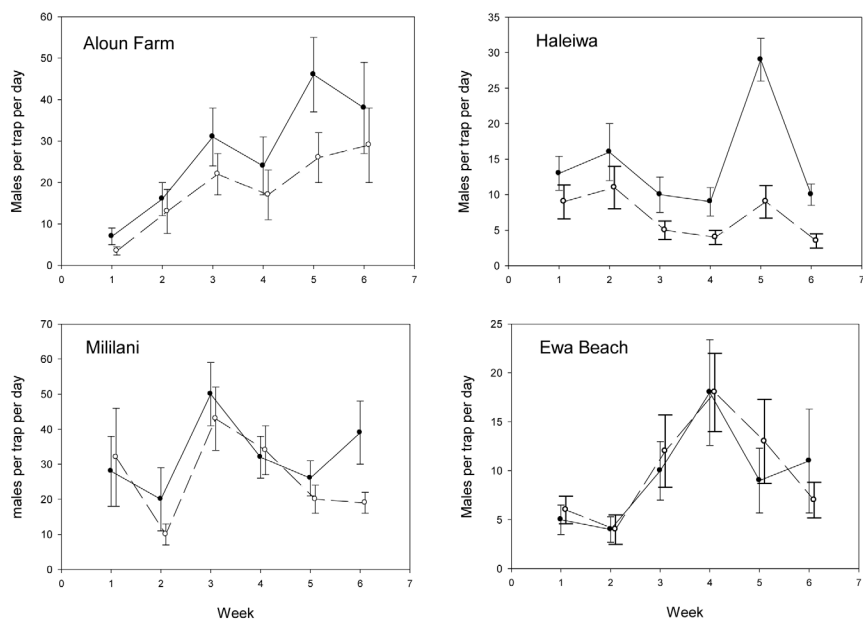


Figure 2. Number of *B. cucurbitae* males captured in Jackson traps baited with cue-lure (CL) liquid (●) versus raspberry ketone formate (RKF) liquid (○) at four study sites on Oahu, Hawaii. At each site, 15 traps of each treatment were operated 1 day per week over 6 consecutive weeks. Symbols represent means (± 1 SE, $n = 15$).

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